Advanced Methods of Pharmacokinetic and Pharmacodynamic Systems Analysis

Volume 2

Edited by
David Z. D'Argenio
University of Southern California
Los Angeles, California

Plenum Press • New York and London

:overy from A3665 than from
it computer simulations cou-
iacokinetic/dynamic studies
)etween new and established
lesign of subsequent Phase 2
new drug.

sagna. Cost of innovation in the

pharmacodynamics and toxicokinetics
pp. 1–13.
r integration of pharmacokinetics,
velopment. *Clin. Pharmacol. Ther.*

III. Mitteilung. *Arch. Psyschiat. Ner-*

III. Mitteilung. *Arch. Psyschiat. Ner-*

d E. Meathe. Pharmacodynamic
:223–40 (1984).
armacokinetics and pharmacody-

D. Verotta and L. B. Sheiner. Phar-
its enantiomers in man. *J. Pharma-*

of narcotic effect: the comparative
y 62:234–41 (1985).
)f the EEG and pharmacodynamic
J. Anaesthesiol. 2:121 (1985).
ween pharmacokinetics and phar-
ity Leiden, The Netherlands, 1991.
lektroencephalographic effects of
troencephalographic effects of mi-
90).
itanski. A new approach to phase
ol. *Anesthesiology* 77:A 370 (1992).
i. The application of pharmacoki-
ment: A-3665 versus fentanyl and

namics, and rational opioid selec-

BUILDING POPULATION PHARMACOKINETIC-PHARMACODYNAMIC MODELS

Jaap W. Mandema

Department of Pharmacy
University of California, San Francisco
and
Department of Anesthesia
Stanford University, School of Medicine

Davide Verotta

Department of Pharmacy
University of California, San Francisco

and
Lewis B. Sheiner

Departments of Pharmacy and Laboratory Medicine
School of Medicine
University of California, San Francisco

INTRODUCTION

One of the major tasks in clinical pharmacology is to estimate population characteristics, such as the mean and inter–individual variability of pharmacokinetic (PK) and pharmacodynamic (PD) parameters in a patient population. Population analysis techniques try to quantify this variability within the population and account for it in terms of patient specific covariates such as age, weight, disease state, concurrent therapy and others. The derived population PK or PD model provides the necessary information required to optimize individual dose: for initial dose, the population mean PK and PD parameters and their relationships with patient specific covariates suffice; for subsequent dose adjustment, the probability distribution of inter–individual random effects which capture inter–individual differences that are not explicable by patient specific covariates; and the residual variability due to measurement errors and intra–individual variability can be used, along with observed individual responses to the initial dose, to arrive at an adjusted dose.

Population analysis according to (nonlinear) mixed effects models, as a means of obtaining population PK–PD parameter estimates, has gained considerable popularity during the past decade because it permits PK–PD characterization of actual patient populations. It permits such characterization because it can make use of only a few samples from each individual collected during routine clinical care or as a supplement to a study designed for other purposes. Certain patient groups,

Advanced Methods of Pharmacokinetic and Pharmacodynamic Systems Analysis, Volume 2, Edited by D.Z. D'Argenio, Plenum Press, New York, 1995

especially the elderly, the critically ill, and children may be hard to study under the rigorous conditions of well controlled clinical trials, and for them, sparse sample based methods may be the only practical way to determine population PK–PD.

The discovery of a population model that adequately describes a given data set is often a complicated and time consuming task. This is partly due to the fact that the relationships of greatest interest, those between PK or PD parameters and covariates, are not directly observed; only the consequences of those relationships, concentrations or effects vs time, are observed. The modeling task consists not only in identifying those covariates (patient specific characteristics) that significantly influence the PK or PD parameters but also consists in characterizing the shape of the relationships between covariates and parameters. In a previous paper [1] we presented some model-building techniques that simplify this task. In this paper we review that previous work, and also extend it. We present here methods that, (i) efficiently find and depict relationships between PK or PD parameters and patient covariates when they exist, (ii) screen for potential interactions between covariates, and (iii) provide diagnostics for detecting individuals that have an unusually large influence on the choice of covariates or shape of relationships. While the NONMEM program [2,3] is used for the work reported here, the ideas and techniques we discuss are general and are easily transferred to other nonlinear mixed effects analysis platforms.

BASIC POPULATION MODEL

The population model has two parts: (i) a model for the response of an individual within the population (individual model) and (ii) a model relating the individual PK–PD parameters to observable patient characteristics (population model).

The jth measurement (e.g., plasma concentration or effect) for the ith individual can be related to the PK or PD parameters in the following way (individual model)

$$y_{ij} = f(\phi_i, x_{ij}) + \epsilon_{ij} \tag{1}$$

where f is a function (PK–PD model) describing the expected value of the response for a given parameter vector ϕ_i as a function of the covariate values x_{ij}. ϕ_i is the vector of the model parameters (such as Cl, EC_{50}, etc.) for the ith individual and x_{ij} is the set of known covariates for this individual (such as dose, time, etc.). The term ϵ_{ij} accounts for the (random) error between the true value and the corresponding measurement. It is generally assumed that ϵ_{ij} are independently normally distributed with either a constant variance σ^2 for all observations (additive residual error model) or with a variance $\sigma^2 f(\phi_i, x_{ij})^2$ that is a function of the predicted drug concentration or effect measurement (proportional residual error model).

An important part of variability in pharmacokinetic and pharmacodynamic experiments is due to differences between patients. The individual PK–PD parameters can be related to observable patient characteristics in the following way (population model)

$$\phi_i = g(\theta, z_i) + \eta_i \tag{2}$$

where g is a known function that describes the expected value of ϕ_i as a function of known individual specific covariates z_i, such as age, weight, disease states, etc., and the vector of population average parameters θ. The vector η_i express the random

difference between the population predictions and individual PK–PD parameters. It is assumed that η_i are independently, and multivariately normally distributed with mean zero and variance–covariance matrix Ω (diagonal elements denoted ω_k^2 where k denotes the kth parameter). Frequently, an exponential inter–individual error model is used which prescribes a log–normal distribution for the PK–PD parameters

$$\phi_i = g(\theta, z_i) \cdot \exp(\eta_i) \tag{3}$$

in which case ω_k is an approximate coefficient of variation.

DATA

Data on four drugs were used to illustrate our model building and evaluation methods (these are the same data sets used in our previous report [1]). The analysis reported here for these drugs are not meant to be definitive but are presented only to illustrate the procedures which are the subject of this paper.

The first drug is ibuprofen, a nonsteroidal anti–inflammatory. Ibuprofen was administered as a single oral dose of 5 mg/kg or 10 mg/kg to 92 children with febrile illness. Two to 6 plasma samples were obtained from each individual up to 8 hours post dosing (total of 411 plasma samples). The following demographic data were recorded: gender (SEX), male (53), female (40); race (RACE), caucasian (17), black (76); location (LOC), clinic (65), hospital/inpatient (28); food since dosing (FED), no (25), yes (68); dose level (DRG), 5 mg/kg (45), 10 mg/kg (48); height (HT), 64–148 cm; weight (WT), 5.6–45.5 kg; age (AGE), 3–132 months. These data were originally reported in [4], and were kindly supplied by Bristol Myers, Hillside, New Jersey.

The second drug is the antiarrhythmic quinidine, administered orally to 136 hospitalized men for various arrhythmias. Plasma samples were obtained for routine clinical purposes (total of 361 samples). The following demographic data were recorded: race (RACE), caucasian (91), latin (35), black (10); smoking (SMO), no (91), yes (45); ethanol abuse (ET), no ethanol abuse or social drinker (90), current ethanol abuse (16), history of ethanol abuse (30); congestive heart failure (HF), no or mild (56), moderate (40), severe (40); creatinine clearance (RF), < 50 ml/min (48), > 50 ml/min (88); weight (WT), 41–119 kg; height (HT), 154–202 cm; age (AGE), 42–92 years; α–1–acid glycoprotein concentration (GLP), 39–316 mg/dl. These data were originally reported in [5], and were kindly supplied by Thomas M. Ludden.

The third drug is the antihypertensive prazosin, administered twice daily in a dose range of 1 mg per day up to 20 mg per day in patients with mild, moderate or severe hypertension. Each patient started with the lowest dose and the dose was increased until the patient's arterial pressure was controlled. After at least a period of eight weeks continuous therapy the PKs of prazosin were studied in 64 patients during a 12 hr dosing interval. After 4 weeks, 35 patients were restudied. A total number of 887 plasma samples were obtained. The following demographic data were recorded: gender (SEX), male (63), female (36); race (RACE), caucasian (67), black (32); visit number (VIS), visit 15 (53), visit 16 (46); smoking (TOB), no (74), yes (25); prior therapy (PT), no (3), yes (96); co–treatment with hydrochlorothiazide (HCTZ), no (44), yes (55); co–treatment with propranolol (PROP), no (85), yes (14); other concomitant therapy (CON), no (24), yes (75); food (FF), fed (50), fasting (49); height (HT), 140–188 cm; weight (WT), 51–139 kg; age (AGE), 24–69 years; serum creatinine (SECR), 0.6–1.8 mg/dl. These data were originally reported in [6], and were kindly supplied by Eli Lilly, Indianapolis, Indiana.

The last drug is the broad spectrum antibiotic pefloxacin. The PK of pefloxacin were studied in 74 critically ill patients with infections with organisms sensitive to pefloxacin. The drug was administered twice daily, 200 mg or 400 mg, in 1 hr infusions. For each individual three plasma samples were obtained during a 12 hr dosing interval. In total 113 dosing intervals were studied (337 plasma concentrations). The following demographic data were recorded: age (AGE), 18–84 years; weight (WT), 43–125 kg; creatinine clearance (CLCR), 0.4–312 mL/min; Glasgow score (GLAS), 3–15; simplified acute physiology score (SAPS), 1–26; albumin (ALB), 17–40 g/L; bilirubin (BIL), 4–150 ämol/L; alanine amino transferase (ALAT), 3–200 IU/L; alkaline phosphatase (AP), 32–615 IU/L; prothrombin level (PT), 36–100 % normal; systolic blood pressure (SPB), 60–175 mmHg; heart rate (HR), 60–170 beats/min; artificial ventilation (AV), no (42), yes (71); gender (SEX), male (86), female (27); center where study was performed (CEN), center 1 (44), center 2 (69); administration of high dose catecholamine (CAT), no (92), yes (21). These data were originally reported in [7], and were kindly supplied by Rhone Poulenc Rorer, Antony, France.

Fig. 1. Time profil
tural PK–model wi
NONMEM to the a
plasma concentratic

MODEL BUILDING PROCEDURE

The population model characterizing the relationships between patient specific characteristics (covariates) and PK–PD parameters is derived using the stepwise approach previously described [1]. Briefly, in a first step individual empirical Bayes estimates of PK–PD parameters are obtained based on a prior NONMEM fit using no covariates. Subsequently, the relationship between the individual PK–PD parameter estimates and candidate covariates is evaluated outside NONMEM using a flexible semi–parametric regression model: the generalized additive model (GAM). Finally, the population analysis is completed using NONMEM on the basis of the GAM analysis of the previous step. This procedure is an extension of the graphical method proposed by Maitre et al. [8]. The model building procedure described above is demonstrated in full in this paper using ibuprofen. The results for the other drugs are used to address specific issues and to evaluate the robustness of the approach.

as data, this step
and we take ad
The relationship
general case be c

Without son
fiable, and even
than two. Furthe
grows exponenti
makes the estim
highly inaccurate

STRUCTURAL PK–PD MODEL

In the first step of the analysis the structural PK–PD model without any covariates is derived and fitted to the data. For ibuprofen, using NONMEM (version 4, level 1.0), the time profile of plasma concentrations is best described using a one–compartment model with first order absorption (ADVAN2, see Fig. 1). The model is parameterized in clearance (Cl), volume of distribution (V) and the first order absorption rate constant ka. An exponential error model is used to characterize the inter–individual variability in the PK parameters. A proportional error model is used for the residual variability. The values of the population parameters θ, σ^2, and Ω are estimated using the the so–called first order method in NONMEM. Subsequently, these parameter values are used to obtain empirical Bayes estimates of the individual PK parameters ϕ_i using the POSTHOC step in NONMEM [9,10].

However, this m
grounds, that a
more general app

COVARIATE MODELS

In the second step a regression model is derived that describes the dependence of the individual PK–PD parameter estimates (i.e., the elements of ϕ_i) on the candidate covariates $z_{1i} \ldots z_{Ni}$. With the estimates of ϕ_i from the Bayes estimation step treated

where $g_n(.)$ is an
appealing becaus
wherever splines
spline with two in
equivalent to 4 de
the interpretation
various covariates

efloxacin. The PK of pefloxacin
ons with organisms sensitive to
200 mg or 400 mg, in 1 hr infu-
e obtained during a 12 hr dosing
337 plasma concentrations). The
\GE), 18–84 years; weight (WT),
.L/min; Glasgow score (GLAS),
-26; albumin (ALB), 17–40 g/L;
sferase (ALAT), 3–200 IU/L; al-
in level (PT), 36–100 % normal;
rate (HR), 60–170 beats/min; ar-
:X), male (86), female (27); center
ıter 2 (69); administration of high
data were originally reported in
)rer, Antony, France.

ionships between patient specific
rs is derived using the stepwise
:t step individual empirical Bayes
on a prior NONMEM fit using no
the individual PK–PD parameter
ıtside NONMEM using a flexible
d additive model (GAM). Finally,
JMEM on the basis of the GAM
extension of the graphical method
ng procedure described above is
ı. The results for the other drugs
the robustness of the approach.

PK–PD model without any covari-
)fen, using NONMEM (version 4,
)ns is best described using a one–
ADVAN2, see Fig. 1). The model
ıstribution (V) and the first order
ror model is used to characterize
:ers. A proportional error model is
population parameters θ, σ^2, and Ω
ethod in NONMEM. Subsequently,
ıl Bayes estimates of the individual
)NMEM [9,10].

ed that describes the dependence of
he elements of ϕ_i) on the candidate
n the Bayes estimation step treated

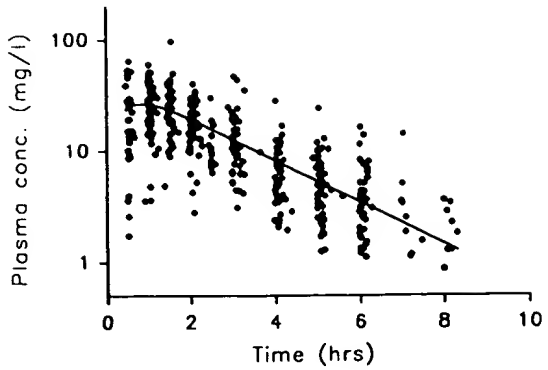


Fig. 1. Time profile of ibuprofen plasma concentrations. The solid line shows the fit of the structural PK–model without any covariates (one–compartment model with first order absorption) using NONMEM to the actual plasma concentrations (•) obtained from 92 children with febrile illness. The plasma concentrations were normalized for a dose of 100 mg.

as data, this step corresponds to the classical regression problem of variable selection and we take advantage of the recent work done on this problem by others [11]. The relationship between covariates and a specific model parameter can in the most general case be described by a multidimensional smooth function $g(..)$

$$\phi_i = g(z_{1i} \ldots z_{Ni}) + \eta_i \tag{4}$$

Without some constraints, such a general description of the data is non–identifiable, and even if identifiable, would be difficult to interpret in dimensions higher than two. Furthermore, the number of points required to fill a space to a given density grows exponentially with the dimension of the space, but usually data do not. This makes the estimation of complex multivariate functions using sparse noisy data highly inaccurate. A multiple linear regression model is an often used simplification

$$\phi_i = \theta_0 + \sum_{n=1}^{N} \theta_n z_{ni} + \eta_i \tag{5}$$

However, this model makes a strong assumption, often unjustifiable on scientific grounds, that a linear relationship exists between parameters and covariates. A more general approach uses a generalized additive model (GAM) [11,12]

$$\phi_i = \theta_0 + \sum_{n=1}^{N} g_n(z_{ni}) + \eta_i \tag{6}$$

where $g_n(.)$ is an arbitrary univariate function. The use of splines for $g_n(.)$ is very appealing because splines are both flexible and convenient to use. (In this paper, wherever splines are mentioned, it is understood that we refer to a natural cubic spline with two internal knots at the 33% and 66% quantiles of the argument variable, equivalent to 4 degrees of freedom [13].) Because of the additive structure of a GAM, the interpretation of the analysis is straightforward, and the independent role of the various covariates can easily be displayed graphically.

Table I. Change in residual sum of squares (ΔRSS) when the individual (posterior Bayes) clearance estimates of ibuprofen are regressed independently on each covariate.

	linear vs. constant		spline vs. linear	
covariate	ΔRSS	p	ΔRSS	p
WT	-3.89	<0.01	-0.10	0.57
AGE	-3.88	<0.01	-0.13	0.49
HT	-3.64	<0.01	-0.21	0.33
FED	-0.61	0.03		
DRG	-0.58	0.03		
RACE	-0.46	0.06		
LOC	-0.07	0.45		
SEX	-0.03	0.60		

INITIAL SCREENING

As a preliminary screening step before the GAM analysis, the individual PK parameter estimates are regressed independently on each covariate. This step is equivalent to the method proposed by Maitre et al. [8] and may give a first impression of the relative importance of each covariate and of the shape of the relationship between it and the model parameters. The results of this screening step for ibuprofen, with respect only to the parameter clearance, are summarized in Table I. The covariate relationships are modeled linearly or with splines. The significance (p) of the linear vs constant model and the spline vs linear model is tested using the F test. The initial screening shows that AGE, WT, HT, DRG, and FED may significantly ($p < 0.05$) influence the clearance of ibuprofen and that the relationship between the continuous covariates and clearance is best described by a linear model.

GENERALIZED ADDITIVE MODEL

The generalized additive model is derived using a step–wise addition/deletion method [12]. This method steps through a series of models according to the following algorithm. At each step of the model building each of the covariates is allowed to be deleted from the model, or to enter the model in any of several prespecified functional representations; e.g., linearly or as a spline. At each step the model is advanced by addition, deletion, or replacement of the single additive term that results in the largest decrease in the Akaike Information Criterion (AIC). (The AIC is proportional to the residual sum of squares from the GAM fit plus a penalty, proportional to the number of parameters in the model.) The search stops when the AIC has reached a minimum value. We estimate the GAM using the statistical programming environment S–plus (version 3.0, Statistical Sciences Inc., Seattle, WA). However, a FORTRAN program to perform the GAM analysis is also available (ref. 11, page 307).

A GAM was derived for all PK parameters of ibuprofen; Cl, V, and ka, however only the results for Cl are shown as an example. For ibuprofen the GAM analysis indicated that Cl is a linear function of WT and a function of the categorical covariates DRG and RACE. The GAM was derived using both a forward search, starting with the NULL model and a backward search, starting with the FULL model, which includes all covariates. A total of 71 different models were tested for Cl. The path taken to the final GAM is summarized in Table II. This table also shows some of the

Table II. The top part o
(Cl) of ibuprofen, star
the largest decrease in
shows some models cl
the notation of the mo
figuratively and not lit

ste
1
2
3
4

Summ

models that (based on th
data. Table II shows that
observed variability in Cl
a small additional chang
6% of the variability in Cl
according to the initial sc
that these two covariates

The relationships bet
shown in Fig. 2. The solid
of each of the terms in the
plotted represents the cov
Cl estimates minus the pr
shown.

NONMEM ANALYSIS

In the final (NONMEI
population model is deriv
Two strategies (roughly c
tion) can be used in the N
all covariates (with associa
the lowest AIC value, and
redundant covariates; or (i
and attempt, thereafter, to
variates appearing in mod
covariates can be tested in
prefer the second approac
large. The parameter esti
for the NONMEM analysis
objective function, –2 log li
reduced model is asympto

Table II. The top part of the table shows the path taken to the final GAM for the clearance (Cl) of ibuprofen, starting with the NULL model. In each step the term that results in the largest decrease in the AIC is added (+) to the model. The bottom part of this table shows some models close to the final GAM (model with smallest AIC value). The ~ in the notation of the models means "is a function of" and the + signs are to be interpreted figuratively and not literally.

step	term	ΔRSS	RSS	AIC
1	constant	12.16	234.3	
2	+WT	-3.89	8.26	200.5
3	+DRG	-0.36	7.90	198.2
4	+RACE	-0.32	7.58	196.3

Summary of model close to the minimum AIC model

Equation	AIC
Cl~WT+DRG+RACE	196.3
Cl~WT+DRG+RACE+LOC	197.3
Cl~WT+DRG+RACE+SEX	197.3

models that (based on the AIC) are very close to the model that best describes the data. Table II shows that WT has the greatest influence on Cl, explaining 32% of the observed variability in Cl. DRG and RACE are not as important, and only result in a small additional change of the residual sum of squares, explaining an additional 6% of the variability in Cl. HT and AGE which appeared to be important covariates according to the initial screening do not appear in the GAM. The reason for this is that these two covariates are highly correlated with WT ($r > 0.940$).

The relationships between covariates and ibuprofen Cl for the final GAM are shown in Fig. 2. The solid lines displayed in this figure are the additive contributions of each of the terms in the GAM, i.e., the $g(.)$ for each of the covariates. The actual data plotted represents the covariate vs partial residual values. The latter is the individual Cl estimates minus the predictions based on all other covariates apart from the one shown.

NONMEM ANALYSIS

In the final (NONMEM) step of the model–building, the non–linear mixed effect population model is derived, based on the regression models found in the GAM step. Two strategies (roughly corresponding to backward elimination and forward addition) can be used in the NONMEM step: (i) include in the initial NONMEM model all covariates (with associated functional relationships) that appear in the GAM with the lowest AIC value, and in models close to it, and attempt, thereafter, to eliminate redundant covariates; or (ii) start with the final GAM as the initial NONMEM model and attempt, thereafter, to add non–redundant covariates, selecting them from covariates appearing in models close to the final GAM (see e.g. Table II). The potential covariates can be tested in the order of their importance in the GAM analysis. We prefer the second approach, especially when the number of candidate covariates is large. The parameter estimates of the GAM analysis are used as initial estimates for the NONMEM analysis. Model selection is done on the basis of the NONMEM objective function, –2 log likelihood (–2LL). The difference in –2LL between a full and reduced model is asymptotically χ^2 distributed with degrees of freedom equal to the

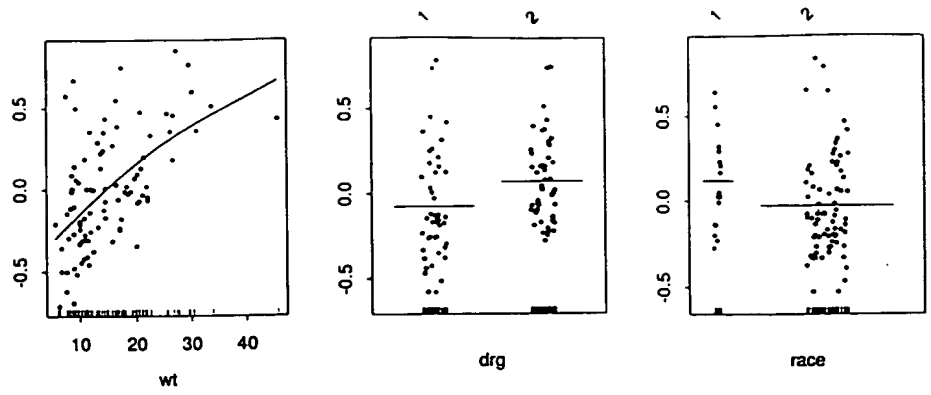


Fig. 2. Partial residual plot of each of the additive terms of the final GAM for the Cl of ibuprofen. The solid line in each plot shows the contribution of a term to the additive predictor of Cl. The actual data in each of the plots are the partial residuals; i.e., the observed individual Cl values minus the prediction based on all terms apart from the one shown. Each curve has been centered to have an average value of zero. The same scale is used for the y–axis in each plot so that the relative importance of the covariates can be compared. The left panel shows the relationship between WT and Cl. The solid line represents the contribution to the GAM, characterized by a natural cubic spline with two internal knots at the 33% and 66% quantiles: 11 kg and 17 kg. The middle panel shows the effect of the two dose level groups (DRG, 1= 5 mg/kg and 2= 10 mg/kg). The right panel shows the effect of race (RACE, 1=caucasian, 2=black). For these categorical covariates the points are randomly shifted by small amounts on the abscissa to display them better.

Table III. Change in –2LL when each of the parameters of the covariates appearing in the final NONMEM model for ibuprofen is set to zero.

Covariate	WT	DRG	RACE
-2ΔLL (vs. θ=0)	115.0	16.0	9.9

difference in number of parameters between the two models. Covariates are added to the model if they significantly decrease the –2LL (> 6.6; p < .01), or deleted from the model if –2LL does not increase significantly. Covariates are also deleted from the model if ± 1 (asymptotic) SE of the parameter estimate includes zero or when the magnitude of the covariate effect is small on clinical grounds.

The NONMEM step in the ibuprofen model building exercise results in the following population model for Cl

$$Cl = (\theta_1 + \theta_2(WT - 15) + \theta_3 DRG + \theta_4 RACE) \cdot \exp(\eta_{Cl}) \quad (7)$$

Table III summarizes the change in –2LL if each of the θ_s in the final NONMEM model is set to zero, showing the significance of each of the parameters. Including the final set of covariates in the NONMEM model resulted in a decrease of the inter–individual variability from 46% to 29% for Cl. No additional covariate results in a significant change in –2LL.

Table III also shows that the GAM analysis not only provided all the important covariates for the final NONMEM model, but also that it correctly identified the relative (order of) importance of each (compare Table III and Table II), and the correct shape of the relationship. To check the latter, the relationship of WT to ibuprofen Cl (the most important relationship) was modeled in NONMEM with a spline. This

Table IV. Summary of covariates to the GAM and NONMEM analy order of appearance in the GAM ibuprofen, 9 for quinidine, 13 for

ANALYSIS	COVARIATES		
Ibuprofen			
GAM	WT	DRG	
NONMEM	WT	DRG	
Quinidine			
GAM	GLP	WT	
NONMEM	GLP	WT	
Prazosin			
GAM	HT	SEX	
NONMEM	HT		
Pefloxacin			
GAM	CICR	BIL	
NONMEM	CICR	BIL	

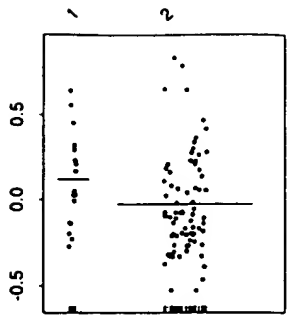
nonparametric representation did firming the results from the GAM from a linear one.

Similar results were found for LOC, and DRG) and their functio final NONMEM model. One add final GAM model was found to sig However, this covariate does appe model.

VALUE OF THE GAM STEP

To decide if the GAM step is does what it is designed to do, i.e., of their functional relationships to simpler approach. Our impression

Regarding achieving its goal, T by the GAM analysis to significantly that are significant in the NONME selects all the important covariates AIC model. The latter case happe model close to the minimum model. of the covariates. The covariates th the GAM analysis also resulted in step. Conversely, those covariates analysis are of minor significance



al GAM for the Cl of ibuprofen.
ditive predictor of Cl. The actual
individual Cl values minus the
ve has been centered to have an
ot so that the relative importance
nship between WT and Cl. The
a natural cubic spline with two
middle panel shows the effect of
ie right panel shows the effect of
the points are randomly shifted

ie covariates appearing in

ACE
9.9

dels. Covariates are added
.6; $p < .01$), or deleted from
iates are also deleted from
e includes zero or when the
inds.
ling exercise results in the

$$\cdot) \cdot \exp(\eta_{Cl}) \qquad (7)$$

θ_i in the final NONMEM
the parameters. Including
d in a decrease of the inter–
tional covariate results in a

provided all the important
t it correctly identified the
nd Table II), and the correct
onship of WT to ibuprofen
NMEM with a spline. This

Table IV. Summary of covariates found to significantly influence clearance according to the GAM and NONMEM analysis for all four drugs. The covariates are listed in their order of appearance in the GAM. The total number of covariates evaluated was 8 for ibuprofen, 9 for quinidine, 13 for prazaosin, and 16 for pefloxacin.

ANALYSIS	COVARIATES							
Ibuprofen								
GAM	WT	DRG	RACE					
NONMEM	WT	DRG	RACE					
Quinidine								
GAM	GLP	WT	RACE	ET				
NONMEM	GLP	WT	RACE	ET	RF			
Prazosin								
GAM	HT	SEX	RACE	TOB	AGE	HCTZ	SECR	WT
NONMEM	HT		RACE		AGE	HCTZ		
Pefloxacin								
GAM	CICR	BIL	WT	AGE	AP	CEN	SEX	SPB
NONMEM	CICR	BIL	WT	AGE		CEN		SPB

nonparametric representation did not result in a significant decrease in –2LL, confirming the results from the GAM analysis that this relationship is indistinguishable from a linear one.

Similar results were found for V. The GAM step provided three covariates (WT, LOC, and DRG) and their functional relationships, all of which are retained in the final NONMEM model. One additional covariate, SEX, that is not present in the final GAM model was found to significantly influence the fit in the NONMEM step. However, this covariate does appear in the GAM model closest to the minimum AIC model.

VALUE OF THE GAM STEP

To decide if the GAM step is worth the trouble, one must consider whether it does what it is designed to do, i.e., identify the important covariates and the shape of their functional relationships to parameters; and whether it does so better than a simpler approach. Our impression is that the answer to both questions is "yes".

Regarding achieving its goal, Table IV compares the covariates that are selected by the GAM analysis to significantly affect the Cl of the four drugs and the covariates that are significant in the NONMEM analysis. For all four, the GAM step either selects all the important covariates or includes them in GAMs close to the minimum AIC model. The latter case happened only once. For quinidine, RF appeared in a model close to the minimum model. The GAM also identifies the relative importance of the covariates. The covariates that provided the greatest decrease in AIC during the GAM analysis also resulted in the largest change in –2LL in the NONMEM step. Conversely, those covariates that are of borderline significance in the GAM analysis are of minor significance in the final NONMEM model. Similar results

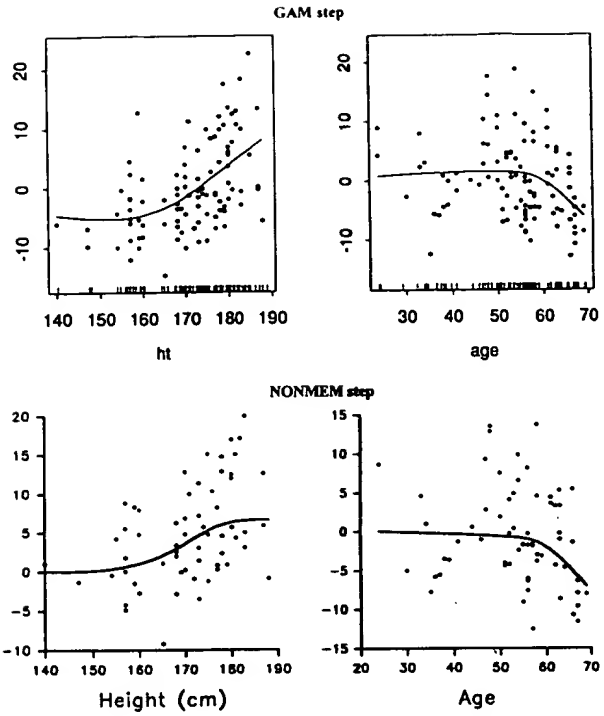


Fig. 3. Partial residual plots for the contribution of HT and AGE to the final GAM for the clearance of prazosin (upper panels) and for the contribution of HT and AGE to the additive NONMEM model (lower panels). The relationships are described by a natural cubic spline with two internal knots at the 33% and 66% quantiles: 168 cm and 177 cm for HT and 52 and 58 years for age. Note that the GAM plots are shifted on the y–axis in comparison to the NONMEM plots because they are centered to have an average of zero. The NONMEM plots show zero effect at the lowest values for HT and AGE. See legend to Figure 2 for further details.

were found for V. So far we have observed that the GAM analysis is inclusive. For all four drugs a total of 27 covariates were found to be of significance in the NONMEM models. Of these covariates, 25 were found in the final GAM. The other 2 were found in models very close to the minimum. Regarding the shape of the functional relationships, prazosin and pefloxacin provide examples. For the former, the GAM analysis identifies important nonlinearities in the relationship between Cl and the covariates HT and AGE. The spline contribution of each of these covariates to the GAM is shown in Fig. 3. A similar nonlinear relationship was included in the NONMEM model, yielding a significant decrease (30.0 points) in –2LL (4 additional parameters) when compared to a linear model for both covariates. The spline relationships found in the NONMEM step are also shown in Fig. 3. Note the similarity between the splines found in the GAM and NONMEM steps. For pefloxacin the GAM analysis indicates a nonlinear relationship between Cl and WT. A similar relationship is found in the NONMEM step, resulting in a significantly better fit than with the linear model.

Regarding whether the GAM analysis provides a better starting point than a simpler approach, we compared it to the one–covariate–at–a–time initial screening step. For pefloxacin 13 out of the 16 covariates are found to significantly affect Cl according to the one–covariate–at–a–time analysis ($p < 0.05$). The GAM analysis

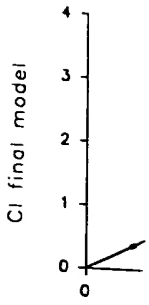


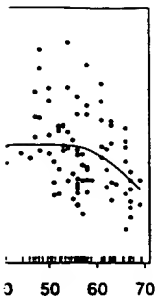
Fig. 4. Plot of empirical Bayes estimates from the basic PK–model without any c all covariate effects (y–axis). The solid li

narrows this down to 8 covariate: analysis. One of the covariates t not selected by the one–covariate Similar results are found for the time analysis tends to select num NONMEM analysis, probably bec of other covariates. Of greater im time analysis disregards some of the final NONMEM model. The r other covariate or by collinearities

These results show that the G initial model for the NONMEM ar derive the GAM model is conside NONMEM and provides a nice gra covariates and model parameters.

DISCREPANCIES BETWEEN GA

There are several factors that may depend on the quality of the ters. These estimates tend to be "sl therefore, fail to reveal important r detected in the NONMEM step, it a here. One would predict that it mi imprecise or few in number. (ii) T model is built, because the prior cl have not found this to be a proble Bayes estimates of ibuprofen Cl ch is changed from that of the basic covariates) to that of the final NON vious problem, this might prove m or sparse. (iii) The individual PK– the GAM analysis, but not in the N the precision of the empirical Bayes ever, we have not found this to be a one individual from day to day, ther



to the final GAM for the clearance
to the additive NONMEM model
spline with two internal knots at
d 58 years for age. Note that the
M plots because they are centered
t at the lowest values for HT and

GAM analysis is inclusive.
to be of significance in the
in the final GAM. The other
Regarding the shape of the
de examples. For the former,
the relationship between Cl
n of each of these covariates
r relationship was included
ase (30.0 points) in -2LL (4
del for both covariates. The
also shown in Fig. 3. Note
and NONMEM steps. For
tionship between Cl and WT.
, resulting in a significantly

better starting point than a
e-at-a-time initial screening
und to significantly affect Cl
< 0.05). The GAM analysis

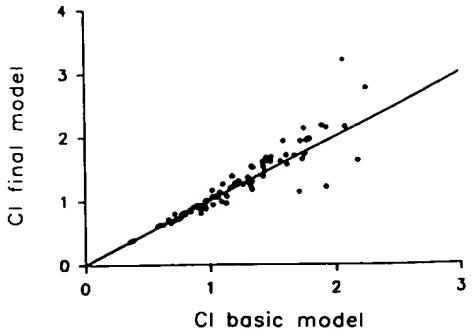


Fig. 4. Plot of empirical Bayes estimates of the individual clearance (Cl) values for ibuprofen obtained from the basic PK–model without any covariates (x–axis) and the final population model including all covariate effects (y–axis). The solid line is the line of unity.

narrows this down to 8 covariates of which 6 are finally selected by the NONMEM analysis. One of the covariates that is selected by the NONMEM analysis, SPB, is not selected by the one–covariate–at–a–time analysis, but does appear in the GAM. Similar results are found for the other drugs. In general, the one–covariate–at–a–time analysis tends to select numerous covariates that are not significant in the final NONMEM analysis, probably because they are collinear with one or a combination of other covariates. Of greater importance, perhaps, is that the one–covariate–at–a–time analysis disregards some of the covariates that appear to be of importance in the final NONMEM model. The reason may be that their effect is masked by some other covariate or by collinearities among covariates.

These results show that the GAM analysis is a robust approach to select a good initial model for the NONMEM analysis. The stepwise addition/deletion method to derive the GAM model is considerably quicker than a similar model search within NONMEM and provides a nice graphical representation of the relationships between covariates and model parameters.

DISCREPANCIES BETWEEN GAM AND NONMEM STEPS

There are several factors that may influence the success of the GAM step: (i) It may depend on the quality of the empirical Bayes estimates of the PK–PD parameters. These estimates tend to be "shrunken" towards the population mean, and may, therefore, fail to reveal important relationships. To the extent that this problem can be detected in the NONMEM step, it appears not to be present in the data sets analyzed here. One would predict that it might appear when individual data are particularly imprecise or few in number. (ii) The empirical Bayes estimates will change as the model is built, because the prior changes. For the four examples presented here, we have not found this to be a problem. Figure 4 shows that the individual posterior Bayes estimates of ibuprofen Cl change only marginally when the prior distribution is changed from that of the basic NONMEM fit (just the structural model without covariates) to that of the final NONMEM fit (including all covariates). As for the previous problem, this might prove more important when individual data are imprecise or sparse. (iii) The individual PK–PD parameter estimates are weighted equally in the GAM analysis, but not in the NONMEM analysis. This could be solved by using the precision of the empirical Bayes estimates as weights in the GAM analysis. However, we have not found this to be a problem; (iv) The covariates may change within one individual from day to day, thereby obscuring covariate–parameter relationships

by "averaging" them within individuals. This can be solved by dividing each individual's data into disjoint contiguous time periods in each of which the covariates are fairly constant. We have applied this technique successfully to the prazosin and pefloxacin data. Clearly, however, the difficulty of trying to detect within–individual correlation between parameters and covariates will be exacerbated as before if data are imprecise or sparse, as now one requires sufficient data in each "period" to allow a reasonably stable empirical Bayes estimate; (v) The AIC is used as the selection criterion in the GAM analysis, whereas the log likelihood test is used in the NONMEM analysis. The AIC tends to select larger models under the operating conditions of our NONMEM step; hence, relative to the latter, the GAM analysis is inclusive, which is an advantage for a screening strategy.

INTERACTION BETWEEN COVARIATES

A disadvantage of the additive structure of the GAM is that interactions between covariates are not included. To take interactions into account, the general structure of a multidimensional smooth function as described by equation (4) could be used. However, this will result in the identifiability problems discussed earlier. Recently, a new approach called PI has become available for covariate selection [14]. This approach automatically selects combinations of covariates showing important interactions and determines their functional relationship using splines. The PI method models the interaction between covariates according to the following equation.

$$\phi_i = \sum_{m=1}^{M} \prod_{n=1}^{N} g_{m,n}(X_{ni}) + \eta_i \qquad (8)$$

where $g_{m,n}(.)$ is a smooth univariate function, in this case a spline. We have used this technique, which is implemented in the FORTRAN program PIMPLE [14], to screen for interactions between covariates in the four data sets, as follows: (i) In a first step, empirical Bayes estimates of η_i were obtained using as prior the model obtained in the NONMEM step. These η_i represent the residual variability in the PK–PD parameters, unexplained by the current additive model; (ii) The best fit of all possible products of two covariates (N in equation 8 equals 2) to the Bayes estimate of η_i for each PK–PD parameter is obtained using the PI method; (iii) The previous NONMEM model is updated by including the candidate interaction determined in the step (ii). The pair of steps (ii)–(iii) can be repeated, first for other two–way interactions, and then for three–way interactions, etc., until no interaction further improves the fit.

For all four drugs, no interactions of this type were found. This suggests that an additive model is often a useful approximation to the more complex true multivariate regression surface. A possible reason is that our data are often not good enough (too noisy and too sparse) to require complex interactions for adequate description.

The PI method can only be used to screen for interactions between continuous covariates. Interactions between categorical covariates and categorical and continuous covariates are therefore analyzed using S–plus. For prazosin an interesting interaction was found. The final additive model in NONMEM showed that Cl is a function of HT, AGE, HCTZ, and RACE and that V is a function of HT and HCTZ. An analysis of the empirical Bayes estimates of η_i using this additive model as a prior showed a profound interaction between HCTZ and SEX for both Cl and V. Figure 5 shows the interaction between SEX and HCTZ for the Cl of prazosin. Addition of a saturated interaction model between HCTZ and SEX to the (additive) NONMEM

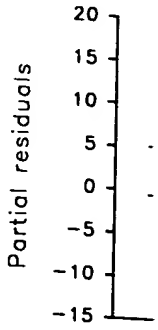


Fig. 5. Partial residual plot showin
prazosin. The partial residuals are p
which indicates that the individuals
mean partial residual values for eac

model results in a significant d
This model can be further refi
not significant and by deleting
resulting, finally, in a model w
with one less parameter. Thu
NONMEM model results in th
that appear to be significant in
probably substituting for the ir

DIAGNOSTICS FOR NONLI

The estimation methods ir
sions of least–squares method
sensitive to "outliers". Thus, r
only a few influential observati
tures of those observations rat
and parameters. It is therefore
an unusual large influence on
focuses on the relationships bet
cific covariates, influence is na
than the level of single observa

In regression analysis, cas
detect influential observations
propose to use two diagnostics
gression analysis: the Cook–sco
case–deletion diagnostics, as w
the data of one individual on
overall influence of the deletion
θ now is used generically to re
and random effect, of the popu
equations:

$$COOK_i = (\backslash$$

solved by dividing each indi-
ı each of which the covariates
ıccessfully to the prazosin and
ng to detect within–individual
e exacerbated as before if data
data in each "period" to allow
AIC is used as the selection cri-
ɹ test is used in the NONMEM
the operating conditions of our
f analysis is inclusive, which is

AM is that interactions between
› account, the general structure
by equation (4) could be used.
ms discussed earlier. Recently,
covariate selection [14]. This
riates showing important inter-
using splines. The PI method
to the following equation.

η_i (8)

case a spline. We have used this
›rogram PIMPLE [14], to screen
ets, as follows: (i) In a first step,
ɜ prior the model obtained in the
ibility in the PK–PD parameters,
ɘst fit of all possible products of
ɜs estimate of η_i for each PK–PD
ɜ previous NONMEM model is
rmined in the step (ii). The pair
–way interactions, and then for
her improves the fit.
ere found. This suggests that an
more complex true multivariate
ı are often not good enough (too
s for adequate description.
nteractions between continuous
ıtes and categorical and contin-
ɹs. For prazosin an interesting
NONMEM showed that Cl is a
is a function of HT and HCTZ.
ng this additive model as a prior
SEX for both Cl and V. Figure 5
the Cl of prazosin. Addition of
ɜEX to the (additive) NONMEM

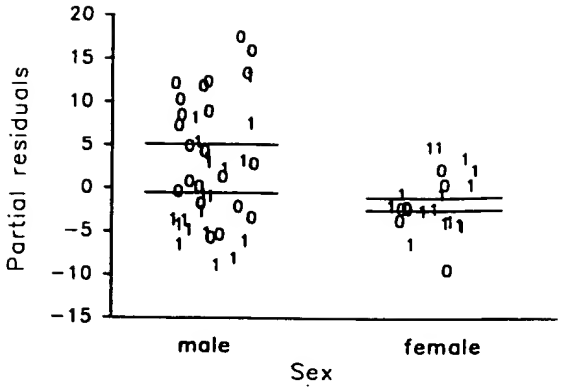


Fig. 5. Partial residual plot showing the interaction between SEX and HCTZ for the clearance of prazosin. The partial residuals are plotted as "0" which indicates no co–treatment with HCTZ or "1" which indicates that the individuals received co–treatment with HCTZ. The solid lines represent the mean partial residual values for each of the four groups.

model results in a significant drop in –2LL of 21.4 on four extra degrees of freedom. This model can be further refined by deleting some of the interaction terms that are not significant and by deleting RACE from the CL model and HT from the V model, resulting, finally, in a model with –2LL 14 points lower than the additive model and with one less parameter. Thus, inclusion of the important interaction term in the NONMEM model results in the removal of two factors, RACE for Cl and HT for V, that appear to be significant in the additive NONMEM model. These two factors are probably substituting for the interaction effect.

DIAGNOSTICS FOR NONLINEAR MIXED EFFECTS MODELS

The estimation methods in NONMEM (and GAM) can be thought of as extensions of least–squares methods. It is well known that such methods can be very sensitive to "outliers". Thus, model building results can be strongly influenced by only a few influential observations, and the fitted model may reflect the unusual features of those observations rather than the general relationship between covariates and parameters. It is therefore very important to detect those observations that have an unusual large influence on the results of the analysis. Since population analysis focuses on the relationships between individual PK–PD parameters and patient specific covariates, influence is naturally defined at the level of the individual, rather than the level of single observations.

In regression analysis, case–deletion diagnostics are the standard tool used to detect influential observations [15,16]. For a non–linear mixed effects model we propose to use two diagnostics that are generalizations of those used in standard regression analysis: the Cook–score (COOK) and covariance–ratio (COV–ratio). These case–deletion diagnostics, as we use them, measure the influence of the removal of the data of one individual on the parameter estimates of the current model. The overall influence of the deletion of the ith individual on the estimate of θ (the symbol θ now is used generically to refer to the vector of all parameters, both fixed effect and random effect, of the population model) can be calculated from the following equations:

$$COOK_i = (\sqrt{(\theta_i - \theta)^T cov(\theta)^{-1} (\theta_i - \theta)}) \quad (9)$$

$$COV - \text{ratio}_i = \sqrt{\frac{\det(cov(\theta_i))}{\det(cov(\theta))}} \tag{10}$$

where θ_i is the vector of parameter estimates when the ith individual is deleted, $cov(.)$ denotes the covariance matrix of the parameter estimates, and $det(.)$ denotes the determinant of a matrix. The Cook–score for the ith individual measures (approximately) the average absolute change in the value of the parameter estimates when the ith individual's data are deleted, each parameter change scaled to its uncertainty (standard error) when estimated using all the data. The COV–ratio for the ith individual measures the "overall" uncertainty (standard error) of all parameter estimates on deletion of the ith individual's data, scaled to the full–data overall uncertainty. A large Cook–score indicates that the individual's data have an inordinate effect on parameter estimate values, and hence are to some degree inconsistent with the rest of the data, while a large value of the COV–ratio indicates that the individual's data contribute considerable information to the fit (are influential). A small value of the Cook–score indicates that the individual's data are compatible with the rest of the data, while a small COV–ratio indicates that parameter precision improves when the individual's data are deleted, suggesting that they are incompatible with the other data.

To detect the influence on a single parameter θ_k of the model, similar diagnostics can be calculated using the following equations

$$COOK_{\theta_{k,i}} = \sqrt{\frac{(\theta_{k,i} - \theta_k)^2}{var(\theta_k)}} \tag{11}$$

$$COV - \text{ratio}_{\theta_{k,i}} = \sqrt{\frac{var(\theta_{k,i})}{var(\theta_k)}} \tag{12}$$

where $var(.)$ denotes the variance of the parameter estimate, and $\theta_{k,i}$ denotes the estimate of θ_k when the data of the ith individual are deleted. The Cook–score for a single parameter and ith individual measures the change in the value of the parameter estimate when the ith individual is deleted, scaled to the uncertainty (standard error) when the parameter is estimated using all the data. The COV–ratio for a single parameter and ith individual measures the uncertainty (standard error) of the parameter estimate on deletion of the ith individual's data, scaled to the uncertainty when estimated from the full–data. The implications of large and small values of these individual parameter diagnostics for individual subjects' data are the same as for the overall diagnostics, except that they are specific for the particular parameter.

The diagnostics presented here are not intended to provide rules for the rejection of data. Influential subjects' data should not necessarily be eliminated from the analysis. These individuals' data may simply be particularly informative. However, influential subjects may point out weaknesses in the analysis and may suggest that the current model is inadequate or may suggest that additional data need to be collected. For example, if the removal of an influential individual changes the value of an estimated parameter to such an extent that its effect becomes insignificant, one is uncomfortable extrapolating the full–data parameter value to future subjects as its value depends, essentially, on just one individual. In general, influential

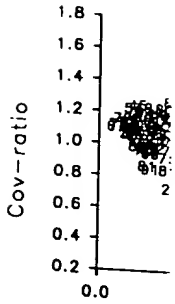


Fig. 6. Plot of the overall Cook–score and the NONMEM model for ibuprofen. The n individuals (up to 92).

individuals are those whose data . (high dimensional) observation–spa with inadequate coverage for reliabl analysis goal [15].

In standard regression analysis. case–deletion diagnostics without tl of each observation (individual). F such methods are currently validate lated by brute force, i.e., the model developed a NONMEM control stre: of the individuals deleted and that timates and corresponding varianc case–deletion diagnostics can then b

EXAMPLE OF CASE-DELETION I

We here present some results o above to examine the ibuprofen ana other 3 drugs.

According to the NONMEM st WT, DRG and RACE (Eq. 7), and V i.

$$V = (\theta_5 + \theta_6(WT - 15) + \theta_7$$

This model was refit to the data with the overall Cook–score and COV–rati 6 identifies certain suspect individua 84 has a particularly low COV–rati (influential?), and numbers 31 and 4 both?). These individuals can be fu COV–ratio for each of the individu: plots for the effect of WT (θ_6), LOC (clearly indicate that the data of indi effect on the value of the estimate o example, individual 47's data chang

(10)

the ith individual is deleted,
r estimates, and $det(.)$ denotes
h individual measures (approx-
the parameter estimates when
change scaled to its uncertainty
The COV–ratio for the ith indi-
error) of all parameter estimates
e full–data overall uncertainty.
ata have an inordinate effect on
egree inconsistent with the rest
icates that the individual's data
fluential). A small value of the
compatible with the rest of the
er precision improves when the
re incompatible with the other

f the model, similar diagnostics

(11)

(12)

r estimate, and $\theta_{k,i}$ denotes the
l are deleted. The Cook–score
s the change in the value of the
leted, scaled to the uncertainty
using all the data. The COV–
sures the uncertainty (standard
h individual's data, scaled to the
implications of large and small
individual subjects' data are the
y are specific for the particular

to provide rules for the rejection
essarily be eliminated from the
ticularly informative. However,
e analysis and may suggest that
that additional data need to be
ial individual changes the value
its effect becomes insignificant,
rameter value to future subjects
ividual. In general, influential

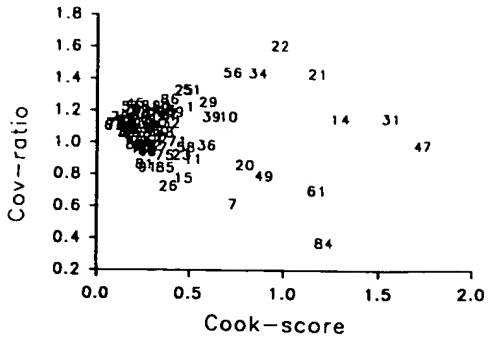


Fig. 6. Plot of the overall Cook–score and COV–ratio when each of the individuals is deleted from the NONMEM model for ibuprofen. The numbers plotted are the actual index number for each of the individuals (up to 92).

individuals are those whose data are far removed from other individuals in the (high dimensional) observation–space; identification of regions of observation space with inadequate coverage for reliable estimation and prediction is an important data analysis goal [15].

In standard regression analysis, numerical methods are available that calculate case–deletion diagnostics without the need to re–estimate parameters after removal of each observation (individual). For nonlinear mixed effect models, however, no such methods are currently validated and case–deletion diagnostics are best calculated by brute force, i.e., the model is refit with each individual deleted. We have developed a NONMEM control stream that automatically refits the model with each of the individuals deleted and that saves the resulting case–deletion parameter estimates and corresponding variance–covariance matrix of the estimates [17]. The case–deletion diagnostics can then be calculated using Eqs. (9)–(12).

EXAMPLE OF CASE-DELETION DIAGNOSTICS

We here present some results of using the case–deletion diagnostics described above to examine the ibuprofen analysis, and also some results of so doing for the other 3 drugs.

According to the NONMEM step of the ibuprofen analysis, Cl is a function of WT, DRG and RACE (Eq. 7), and V is a function of WT, LOC, DRG, and SEX:

$$V = (\theta_5 + \theta_6(WT - 15) + \theta_7 LOC + \theta_8 DRG + \theta_9 SEX) \cdot \exp(\eta_v) \qquad (13)$$

This model was refit to the data with each of the individuals deleted. The values of the overall Cook–score and COV–ratio are shown in Fig. 6 for each individual. Figure 6 identifies certain suspect individuals (those appearing along its "edges"): number 84 has a particularly low COV–ratio (outlier?), number 22 has a high COV–ratio (influential?), and numbers 31 and 47 have high Cook–scores (outlier? influential? both?). These individuals can be further evaluated by plotting the Cook–score vs COV–ratio for each of the individual parameter estimates. Figure 7 shows these plots for the effect of WT (θ_6), LOC (θ_7), DRG (θ_8), and SEX (θ_9) on V. These figures clearly indicate that the data of individuals 31, 84, and 47 have an unusually great effect on the value of the estimate of V when compared to other individuals. For example, individual 47's data changes θ_9 almost 1.5 times the standard error of the

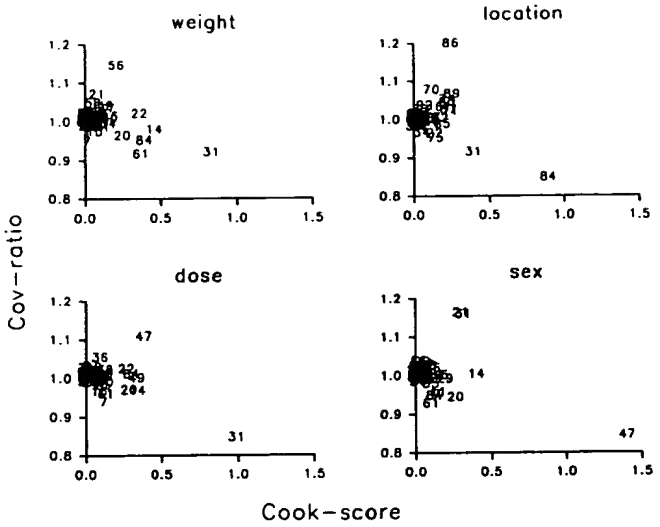


Fig. 7. Plot of the Cook–score and COV–ratio for the parameters describing the effect of WT (θ_6), LOC (θ_7), DRG (θ_8), and SEX (θ_9) on the volume of distribution of ibuprofen. The numbers plotted are the actual index number for each of the individuals (up to 92).

parameter estimate. Table V summarizes the parameter estimates for the model for V when each of these individuals is deleted from the analysis. Removal of individual 31's data renders the effect of DRG insignificant, whereas removal of individual 47's data renders the effect of SEX insignificant. Both effects are highly significant ($p<0.01$) according to the likelihood ratio test when all individuals are included in the analysis. Furthermore, the magnitude of these (binary) effects in the full–data analysis is notable: both change the average V by about 18%. The number of males/females was 53/40, while the number of subjects with low/high DRG was 45/48. Since all these numbers are appreciable, the fact that the apparent effects of the covariates change so profoundly upon deletion of just one individual in each case suggests that the "covariate effect" is not real, but simply an artefact due to outlying individuals. If, however, there had been severe covariate imbalance; i.e., only a few individuals with the same SEX value as number 47, or only a few individuals with the same DRG value as number 31, then one might suspect that the effect could be real, and seek confirmation through study of additional individuals with like covariate values. The effect of removal of the data of individual 84 on θ_7 is also profound. LOC is a highly significant factor when the data of all individuals are analyzed, changing –2LL by 50 points when included in the model. Yet one must conclude that the magnitude of its effect is poorly defined, since it changes by almost 1 standard error when subject 84 is removed. In all but the last of the above cases, it is true that the standard errors of the parameter estimates based on the full data are sufficiently large that the covariate effects are suspect, but the case–deletion diagnostics add considerable specificity to this vague suspicion.

Case–deletion diagnostics will identify influential individuals even though they be otherwise difficult to identify due to the complexity of the population data. Even may plots of the empirical Bayes estimates of the model parameters vs the covariates such as in Fig. 2 may not clearly identify influential subjects because of the complex influence structure of each subject on all parameter estimates.

Case–deletion diagnostics are also helpful in explaining some of the prazosin results. For this drug, several covariates are found to significantly influence Cl (see Table IV) and V, resulting in a marked drop in –2LL when they are included. However,

Table V. Effect of removal of cer
estimates of the NONMEM popu
fen compared to the parameter
figures in each cell are the param
in –2LL (based on the analysis w
set to zero is also shown (first col

	–2ΔLL (vs. θ=0)
θ_5	
θ_6(wt)	77.5
θ_7 (loc)	50.2
θ_8 (drg)	8.3
θ_9 (sex)	14.4
$\omega_V 2$	

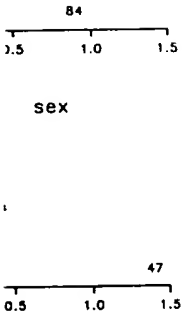
the inter–individual variability i
explanatory covariates, a perple
variability is being "explained", v
deletion diagnostics reveal that t
results in a 25% decrease of inter–
standard error of this estimate. S
individual variability in V. This su
compared to other subjects. How
and V were not well explained by

CONCLUSIONS

In this paper we have shov
approach to selecting a good ini
The GAM step selects the most ir
linearities in the relationships betv
method is efficient and provides il
structure of the GAM is that inte
we here present a screening metho
our four data sets, no interactions
adequate approximation to an ine
surface.

We have also shown that case
data is unusually influential in the
should allow population modelin

location



escribing the effect of WT (θ_6), LOC
ofen. The numbers plotted are the

eter estimates for the model
n the analysis. Removal of
ificant, whereas removal of
cant. Both effects are highly
est when all individuals are
e of these (binary) effects in
erage V by about 18%. The
er of subjects with low/high
ble, the fact that the apparent
eletion of just one individual
t real, but simply an artefact
n severe covariate imbalance;
as number 47, or only a few
en one might suspect that the
udy of additional individuals
ie data of individual 84 on θ_7
ien the data of all individuals
uded in the model. Yet one
y defined, since it changes by
In all but the last of the above
er estimates based on the full
suspect, but the case–deletion
suspicion.
individuals even though they
of the population data. Even
l parameters vs the covariates
bjects because of the complex
timates.
plaining some of the prazosin
significantly influence Cl (see
n they are included. However,

Table V. Effect of removal of certain individuals' data (#31, #47, #84) on the parameter estimates of the NONMEM population model for the volume of distribution of ibuprofen compared to the parameter estimates based on all individuals' data (all #). The figures in each cell are the parameter estimate (standard error of estimate). The change in –2LL (based on the analysis with all #) when each of the parameters estimates (θ) is set to zero is also shown (first column).

	–2ΔLL (vs. θ=0)	all #	- #31	- #47	- #84
θ_5		2.12 (0.16)	2.28 (0.16)	2.19 (0.11)	2.21 (0.14)
θ_6(wt)	77.5	0.111 (0.015)	0.099 (0.014)	0.109 (0.016)	0.117 (0.014)
θ_7 (loc)	50.2	1.91 (0.47)	1.73 (0.42)	1.96 (0.46)	1.50 (0.39)
θ_8 (drg)	8.3	0.38 (0.22)	0.16 (0.19)	0.30 (0.24)	0.31 (0.22)
θ_9 (sex)	14.4	0.37 (0.15)	0.33 (0.18)	0.15 (0.13)	0.35 (0.22)
$\omega_V 2$		0.083 (0.029)	0.094 (0.030)	0.087 (0.029)	0.072 (0.025)

the inter–individual variability in Cl and V is not reduced by inclusion of these explanatory covariates, a perplexing contradiction (unexplained inter–individual variability is being "explained", why then does its magnitude not decrease?). Case–deletion diagnostics reveal that this is due to one subject. Removal of this subject results in a 25% decrease of inter–individual variance in Cl and a 50% decrease in the standard error of this estimate. Similar results are found for the estimate of inter–individual variability in V. This subject did not have unusual Cl and V estimates when compared to other subjects. However, in contrast to the others, this individual's Cl and V were not well explained by the final population model.

CONCLUSIONS

In this paper we have shown that a preliminary GAM analysis is a robust approach to selecting a good initial model for a subsequent NONMEM analysis. The GAM step selects the most important covariates and indicates important non–linearities in the relationships between covariates and PK–PD model parameters. The method is efficient and provides illustrative graphics. A disadvantage of the additive structure of the GAM is that interactions between covariates are not included, but we here present a screening method to detect such interactions. When it is applied to our four data sets, no interactions are found, suggesting that a GAM may often be an adequate approximation to an inevitably more complex true multivariate regression surface.

We have also shown that case–deletion diagnostics can detect individuals whose data is unusually influential in the final NONMEM analysis. Use of such diagnostics should allow population modeling to become more robust.

REFERENCES

1. J. W. Mandema, D. Verotta, and L. B. Sheiner. Building population pharmacokinetic–pharmacodynamic models. I. models for covariate effects. *J. Pharmacokinet. Biop.* 20:511–528 (1992).
2. S. L. Beal and L. B. Sheiner. *NONMEM User's Guides – Part I–VII. NONMEM project group,* University of California, San Francisco, 1992.
3. L. B. Sheiner and T. M. Ludden. Population pharmacokinetics/dynamics. *Annu. Rev. Pharmacol.* 32:185–209 (1992).
4. R. D. Brown, J. T. Wilson, G. L. Kearns, V. F. Eichler, V. A. Johnson and K. M. Bertrand. Single–dose pharmacokinetics of ibuprofen and acetaminophen in febrile children. *J. Clin. Pharmacol.* 32:231–41 (1992).
5. C. N. Verme, T. M. Ludden, W. A. Clementi, and S. C. Harris. Pharmacokinetics of quinidine in male patients: a population analysis. *Clin. Pharmacokinet.* 22:468–480 (1992).
6. M. R. Goldberg, C. S. Sushak, R. W. Rockhold, L. W. Thompson, and the Pinacidil vs. Parazosin Multicenter Investigator Group. Vasodilator monotherapy in the treatment of hypertension: Comparative efficacy and safety of pinacidil, a potassium channel opener, and prazosin. *Clin. Pharmacol. Ther.* 7:78–92 (1988).
7. R. Bruno, M-C. Iliadis, B. Lacarelle, V. Cosson, J. W. Mandema, Y. Le Roux, G. Montay, A. Durand, M. Ballereau, M. Alasia, J. Albanese, G. Francois, A Iliadis, and A. Frydman. Evaluation of Bayesian estimation in comparison to NONMEM for population pharmacokinetic data analysis: application to pefloxacin in intensive care unit patients. *J. Pharmacokin. Biop.* 20:653–669 (1992).
8. P. O. Maitre, M. Bührer, D. Thomson and D. R. Stanski. A three–step approach combining Bayesian regression and NONMEM population analysis: application to midazolam. *J. Pharmacokinet. Biop.* 19:377–384 (1991).
9. L. B. Sheiner and S. Beal. Bayesian individualization of pharmacokinetics: simple implementation and comparison with non–bayesian methods. *J. Pharm. Sci.* 71:1344–1348 (1982).
10. L. B. Sheiner, S. Beal, B. Rosenberg and V. V. Marathe. Forecasting individual pharmacokinetics. *Clin. Pharmacol. Ther.* 26:294–305 (1979).
11. T. J. Hastie and R. J. Tibshirani. *Generalized Additive Models.* Chapman and Hall, New York, 1990.
12. T. J. Hastie. Generalized additive models. In: J. M. Chamber and T. J. Hastie (eds.), *Statistical models* in S. Wadsworth and Brooks, Pacific Grove, California, pp. 249–304, 1991.
13. B–spline package can be obtained from D. Verotta, Box 0446, School of Pharmacy, University of California, San Francisco, CA 94143–0446 upon request (email: david@c255.ucsf.edu).
14. L. Breiman. The PI method for estimating multivariate functions from noisy data. *Technometrics* 33:125–160 (1991).
15. R. D. Cook and S. Weisberg. *Residuals and influence in regression,* Chapman and Hall, New York, 1982.
16. R. Christensen, L. M. Pearson, and W. Johnson. Case-deletion diagnostics for mixed models. *Technometrics* 34:38–45 (1992).
17. NONMEM routines can be obtained from J.W. Mandema, Department of Anesthesia, Stanford University Medical Center, 300 Pasteur Drive, Stanford, CA 94306–5115 upon request (email: jaap@leland.stanford.edu).

MOI